# A new species of *Knipowitschia* (Gobiidae) from Dalmatia, Croatia

by

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**ABSTRACT**. - *Knipowitschia radovici* sp. nov. is described from the Neretva drainage basin, Dalmatia, Croatia. The new species is assigned to the Gobiidae *Knipowitschia* based on diagnostic features belonging to the genus. The new species differs from congeners by the reduction of cephalic lateral line canals, the reduction in squamation and the arrangement of some sensory papillae rows. Morphological characters of *Knipowitschia* species are discussed.

**RÉSUMÉ**. - Une nouvelle espèce de *Knipowitschia* (Gobiidae) provenant de Dalmatie, Croatie.

Knipowitschia radovici sp. nov. est décrite du bassin hydrographique de Neretva, Dalmatie, Croatie. Cette nouvelle espèce est attribuée au Gobiidae Knipowitschia sur la base des caractéristiques diagnostiques du genre. Le degré de réduction des canaux de la ligne latérale céphalique et l'écaillure réduite sont des caractères uniques pour le genre Knipowitschia. Par ailleurs, la position de certaines rangées de papilles sensorielles distingue clairement cette espèce de toutes les autres du genre. Les caractères morphologiques de cette nouvelle espèce de Knipowitschia sont discutés.

Key words. - Gobiidae - Knipowitschia - MED - Adriatic - Systematics - New species.

Several *Knipowitschia* species or subspecies were described during the last decade of the 20th century (Ahnelt and Bianco, 1990; Ahnelt, 1991, 1995; Mrakovčić et al., 1996) and the knowledge on morphology and taxonomy of some species has increased greatly (Economidis and Miller, 1990; Ahnelt et al., 1995). However, only two papers on the taxonomy of genus Knipowitschia in the Adriatic catchment were published recently (Mrakovčić et al., 1996; Kovačić and Pallaoro, 2003), although it was suspected new undescribed freshwater gobiid species exist in this region (Mrakovčić et al., 1996). A sample of unknown Knipowitschia species was collected in the Norin river, a tributary of the river Neretva (43°04'48"N, 17°38'00"E) during the gobiological field work along the eastern Adriatic rivers and lakes in the summer of 1999. The collected specimens represent a new species that is described below.

#### **METHODS**

Morphometric and meristic methods as in Miller (1988). Fin abbreviations: A, anal fin; C, caudal fin; D1, D2, first and second dorsal fins; P, pectoral fin; V, pelvic disc. Morphometric abbreviations: Ab, anal fin base; Ad and Aw, body depth and width at anal fin origin; Cl, caudal fin length; CHd, cheek depth; CP and CPd, caudal peduncle length and depth; D1b and D2b, first and second dorsal fin base; E, eye diameter; H and Hw, head length and width; I, interorbital width; Pl pectoral fin length; PO, postorbital length; SL, standard

length; SN, snout length; SN/A and SN/AN, distance from snout to vertical of anal fin origin and anus; SN/D1 and SN/ D2, distance from snout to origin of first and second dorsal fins; SN/V, distance from snout to vertical of pelvic fin origin; V/AN, distance from pelvic fin origin to anus; Vd, body depth at pelvic fin origin; VI, pelvic fin length. The terminology of the lateral-line system follows Sanzo (1911) and Economidis and Miller (1990). Paratype PMR VP1212 was blanched in a mixture of 9 parts of 0.5% KOH solution and 1 part of 30% H<sub>2</sub>O<sub>2</sub> for three hours and then stained in 2% KMnO4 solution for 20 s and 0.3% H<sub>2</sub>SO<sub>4</sub> solution for 20 s for better examination of sensory papillae rows. The morphological data on Mediterranean, Black, Caspian and Aral Seas *Knipowitschia* species were compared for differential diagnosis (Georghiev, 1966; Miller, 1972; Pinchuk, 1978; Gandolfi et al., 1985; Economidis and Miller, 1990; Ahnelt and Bianco, 1990; Ahnelt, 1991, 1995; Kovačić and Pallaoro, 2003).

## KNIPOWITSCHIA RADOVICI SP. NOV.

(Figs 1, 2)

This species is placed in *Knipowitschia* Iljin, 1927 (type species: *Gobius longecaudatus* var. a and var. b., Kessler, 1877), based on a description of the genus *Knipowitschia* by Economidis and Miller (1990), by its possession of (1) head, back to at least the origin of D2 and abdomen naked, (2) anterior oculoscapular head lateral-line canal ending anteri-

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Figure 1. - *Knipowitschia radovici* sp. nov., holotype, male, 27.9+6.5 mm, PMR VP1211. Scale bar = 5 mm.

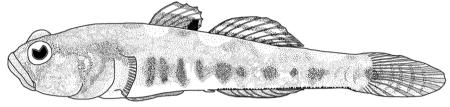


Figure 2. - *Knipowitschia radovici* sp. nov., holotype, male, 27.9+6.5 mm, PMR VP1211, in lateral view. Scale bar = 5 mm.

orly between eyes without pores  $\sigma$ , (3) anterior nostril a short tube, without rim tentacle, (4) pectoral fin with upper rays enclosed by fin membrane, (5) pelvic disc without reduction and pelvic anterior membrane well developed, (6) branchiostegal membrane attached to entire lateral margin of isthmus, (7) sensory papillae with suborbital series a with more than one transverse rows, (8) series c in several transverse rows, (9) row o absent, (10) interorbit without multiple transverse rows of papillae.

## Material

Holotype (Figs 1, 2). - Male, 27.9 + 6.5 mm, PMR VP1211, Vid, Norin river, Croatia, 31 July 1999, leg. M. Kovačić.

Paratypes. - One male, 27.2 + 6.6 mm, PMR VP1212 and one juvenile, 17.8 + 4.4 mm, PMR VP1213, Vid, Norin river, Neretva catchment, Croatia, 31 July 1999, leg. M. Kovačić.

The holotype and paratypes are deposited at the Prirodoslovni muzej Rijeka (PMR).

## **Diagnosis**

Knipowitschia radovici differs from all other species of the genus by the following combination of characters: (1) squamation reduced to axillar and caudal peduncle patches, unconnected or connected with single row along lateral midline, (2) the anterior oculoscapular canal present, ending anteriorly with or without pore  $\lambda$ , (3) the preopercular canal present, at least in a part of population, (4) pelvic frenum with crenate rear edge, (5) suborbital longitudinal row b anteriorly beginning below rear border of eye, (6) anterior dorsal row m present, (7) oculoscapular row u absent,

(8) single papilla of interorbital row p present, at least in a part of population.

# Description

Morphology

Body proportions are given in table I. Body moderately elongate, laterally compressed at caudal peduncle, head large and moderately depressed (Fig. 3, Tab. I). Snout oblique, eyes dorsolateral, eye contour above snout and predorsal area in profile, anterior nostril short, tubular, erect, without process from rim, posterior nostril pore-like, near orbit. Mouth oblique, jaws subequal, posterior angle of jaws below anterior end of pupil. Branchiostegal membrane attached along entire lateral margin of isthmus.

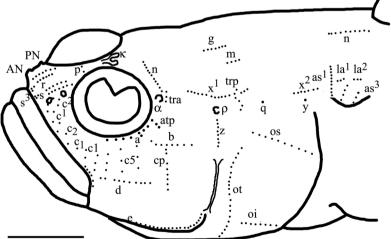


Figure 3. - Knipowitschia radovici sp. nov., head lateral-line sensory papillae and canal pores of paratype, male, 27.2+6.6 mm, PMR VP1212. AN, PN, anterior and posterior nostrils; see other terminology in text. Scale = 2 mm. [Knipowitschia radovici sp. nov., papilles sensorielles de la ligne latérale céphalique et pores des canaux du paratype, mâle, 27.2+6.6 mm, PMR VP1212. AN, PN, narines antérieures et postérieures; voir autre terminologie dans le texte. Échelle = 2 mm.]

Table I. - Body proportions of *Knipowitschia radovici* sp. nov. Values are individual; see abbreviations in methods. [Proportions du corps de Knipowitschia radovici sp.nov. Les valeurs sont individuelles; voir les abréviations dans les méthodes.]

	Adult males		Juvenile
	Holotype PMR	Paratype PMR	Paratype PMR
	VP1211	VP1212	VP1213
SL (mm)	27.9	27.2	17.8
%SL, H	31.2	30.9	33.7
Hw	21.9	24.6	21.3
SN/D1	38.4	38.6	42.1
SN/D2	57.0	56.6	60.7
SN/AN	53.4	53.7	59.0
SN/A	60.6	59.6	61.8
SN/V	31.2	29.8	34.8
CP	24.0	24.3	26.4
D1b	13.6	15.4	12.4
D2b	18.6	18.0	16.3
Ab	15.8	15.8	12.4
Cl	23.3	24.3	24.7
Pl	24.7	25.0	23.6
Vl	21.1	22.8	22.5
Vd	20.8	22.4	21.3
Ad	15.8	16.5	14.6
Aw	11.5	11.4	11.2
CPd	9.7	10.7	10.1
V/AN	22.2	23.9	24.2
%CP, CPd	41.5	39.7	36.7
%H,SN	24.1	23.8	25.0
E	23.0	23.8	26.7
PO	52.9	52.4	48.3
CHd	16.1	17.9	11.7
Hw	69.3	77.9	46.9
%E, I	36.8	40.0	18.8
%V/AN, VI	95.2	95.4	97.6

## Fins

D1 VI; D2 I,8; A I,8; C 12-13 branched rays (12: 1; 13: 2), 16 segmented rays (16: 3); P 15-16 (both sides: 15 and 15: 2, 16 and 16: 1), V I/5 + 5/I. Fin bases and lengths in proportion to standard body length are given in table I. D1 spines III-V extending when depressed to D2 I in adult males, but not reaching D2 in juvenile. Interdorsal space distinct. D2 commences over vertical of anterior beginning of urogenital papilla, with last ray over vertical of last or penultimate A ray. A commences below second segmented ray of D2, with rear tip reaching 1/2 of length of caudal peduncle in adult males, but only 1/4 of length of caudal peduncle in juveniles. C rounded. P rounded, extends back to vertical of D2 I. V rounded, not reaching anus, with frenum in midline about 1/2 length of pelvic spinous ray, with crenate rear edge.

#### Scales

Body with ctenoid scales. Adult males and the juvenile show similar development of squamation: squamation reduced to axillar and caudal peduncle patches, unconnected (holotype and juvenile paratype) or connected with singular row along lateral midline (adult paratype). Axillary patch transversely with four to five rows of scales, longitudinally from axilla to vertical of rear end of D1. Caudal peduncle patch with scales around caudal peduncle, at least in a posterior part, and a single row along lateral midline to the below interdorsal space or just below D2.

## Lateral line system

Head canals. Adult males and the juvenile with similar development of head canals: the anterior oculoscapular canal always present, carrying pores  $\kappa$ ,  $\alpha$ ,  $\rho$ , ending anteriorly with pores  $\lambda$  (holotype) or as open furrows beginning at double  $\kappa$  (paratypes). The preopercular canal developed, carrying pores  $\gamma$  and  $\epsilon$  or just as open furrow. The posterior oculoscapular canal absent.

Rows and the number of sensory papillae (Fig. 3): (I) preorbital: snout with three rows in median preorbital series: internal row r (3-6), outer row s (7-8), anterior row  $s^3$  (3-4). Lateral series c in four parts: superior  $(c^2)$  close to PN (1-2); middle  $c^{1}$  (2-3) close to AN; inferior upper  $c_{2}$  (3-4) and lower  $c_1$  (2-3) above lips. (II) *suborbital*: Infraorbital row a extending forwards from the rear border of eye to anterior half of pupil, consisting of 8-10 longitudinally arranged papillae and a longer transverse row atp (2-4). The transverse proliferation of row a consists of atp and one to three doubled or tripled papillae. Longitudinal row b (6-11) anteriorly beginning below rear border of eye. Six transverse row c below horizontal of row b: c1 (2-3), c2 (3), c3 (3), c4 (3-5), c5 (3-5) and cp (5-9). Transverse row cp at vertical of row tra, beginning below row b, ending below level of row d. Longitudinal row d (14-19) with separated supralabial and posterior part, not reaching posteriorly row cp. (III) preoperculo-mandibular: external row e and internal row i divided into anterior (e: 12-14, *i*: 18-23), and posterior sections (*e*: 14-15, *i*: 21-24); mental row f(3). (IV) oculoscapular: anterior transverse series tra (3) behind pore  $\alpha$ , anterior longitudinal row  $x^{l}$ divided by posterior transverse row, trp (5-8) in anterior section (8-12) above row z and posterior section (3-4), posterior longitudinal row  $x^2$  developed (4-7); row y visible as single papilla in paratypes; row z (5-8) developed, row q (1) behind transverse row trp. Axillary rows  $as^{1}$  (6-8),  $as^{2}$  (3-7),  $as^{3}$ (4-8),  $la^{1}(2-3)$  and  $la^{2}(2-3)$  present. (V) opercular: transverse row ot (18-20); superior longitudinal row os (10-12); and inferior longitudinal row oi (8-12). (VI) anterior dorsal: row *n* (7-11), row *g* (7-8), row *m* (4-5) and row *h* (11-12) present, row o absent. (VII) interorbital: row p (1) present in paratypes. The juvenile (SL = 17.8 mm) matches adults in the presence and position of rows of sensory papillae, but

has smaller number of sensory papillae in some rows (atp, b, d,  $x^{I}$ ,  $as^{2}$ ,  $as^{3}$ ).

## Coloration

In life (from slides of anaesthetized specimens). Adult males (SL > 27 mm): upper part of body ochre. Lateral part between pale saddles and underside greyish brown, with slightly darker blotches, underside whitish to yellowish. Colours in life of juvenile (SL = 17.8 mm) similar with adults, except for a distinct dark mark on caudal fin origin, dark dots along origin of A and ventral midline of caudal peduncle, and belly transparent, with silvery peritoneum visible. The pattern of skin pigmentation in adults and the juvenile is the same as in the preserved specimens and it is described in detail for preserved specimens.

Preserved specimens. Adult males (SL > 27 mm): Upper part of body with four more or less distinctive pale saddles. Underside, including breast and belly, dusty, except darker dots along origin of A and ventral midline of caudal peduncle, belly whitish. Lateral part between pale saddles and underside pale fawn with dark blotches. Scaled areas more intensively pigmented than scaleless. Three to four vertically elongated dark blotches below D1, with height restricted to less than a half of the body depth. Five to six oval dark blotches below D2 and on caudal peduncle. Head pigmented, a bit more densely on opercle and between eye and upper lip. D1 with one lower broad band, second narrow band along the end of the fin, and an intensive spot between lower part of D1 V, VI and rear end of fin; D2 with three pigmented longitudinal bands; C with several vertical curved bands, the rest of the fin uniformly pigmented; P, A and V pigmented. Juvenile (SL = 17.8 mm): Upper part of body pigmented. Three vertically elongated dark blotches below D1 along lateral midline and six oval dark blotches below D2 and on caudal peduncle along lateral midline. Underside whitish, except darker dots along origin of A and ventral midline of caudal peduncle. Head pigmented, a bit more densely between eye and upper lip. D1, D2 and P moderately dotted, on D1 visible developing rear spot. C showing large mark connected with last caudal peduncle blotch. V and A poorly dotted.

#### **Etymology**

The new species is named after my friend ornithologist Dragan Radović, who encouraged and helped me to collect samples along the Adriatic rivers and lakes.

## **Ecology**

*K. radovici* was collected in a karstic river with fresh (0.09 PSU, D. Radović, personal communication), oligotrophic, running water (monthly average 3.5-6.0 m/s<sup>3</sup> during summer, D. Radović, pers. com.). The adults were col-

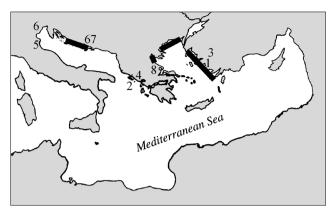


Figure 4. - Geographical distribution of *Knipowitschia* species in the Mediterranean area. (**a**) *K. caucasica*, (1) *K. ephesi*, (2) *K. goerneri*, (3) *K. mermere*, (4) *K. milleri*, (5) *K. panizzae*, (6) *K. punctatissima*, (7) *K. radovici* sp. nov., (8) *K. thessala*. [Distribution géographique de l'espèce Knipowitschia en Méditerranée.]

lected only in the main current at the greatest river depth (5.5 m) by SCUBA diving. The bottom was clear gravel laterally becoming muddy. Juvenile was collected in shallow waters, amongst vegetation, by a handnet from the surface together with *Knipowitschia punctatissima croatica*.

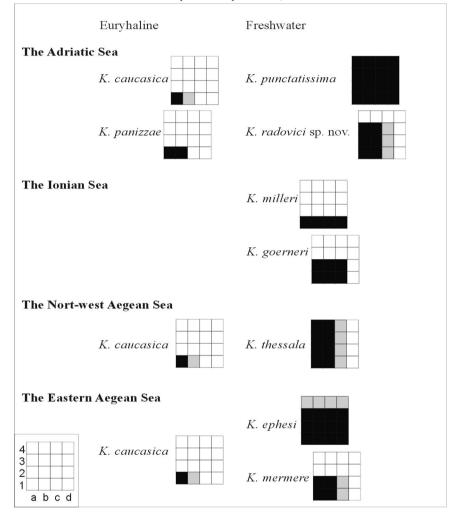
## Geographical distribution

*K. radovici* is only known from the type locality at 10.8 km long Norin river in the village of Vid, the Neretva drainage basin, Dalmatia, Croatia (43°04'48''N, 17°38'00''E) (Fig. 4). The type locality in the village of Vid is situated 2.8 km down river from karstic spring.

# DISCUSSION

Nine species of Knipowitschia genus are known in the Mediterranean area, including the presently described species: K. caucasica (Kawrajsky in Berg, 1916), K. ephesi Ahnelt, 1995, K. goerneri Ahnelt, 1991, K. mermere Ahnelt, 1995, K. milleri (Ahnelt & Bianco, 1990), K. panizzae (Verga, 1841), K. punctatissima (Canestrini, 1864), K. radovici sp. nov. and K. thessala (Vinciguerra, 1921) (Miller, 1972; Gandolfi et al., 1985; Economidis and Miller, 1990; Ahnelt and Bianco, 1990; Ahnelt, 1991, 1995; Kovačić and Pallaoro, 2003) (Tab. II). Seven of them are freshwater fishes, while two others are euryhaline species, occurring from freshwater to brackish water (Miller, 1972: K. panizzae), or even hypersaline water (Ahnelt et al., 1995: K. caucasica). K. caucasica is a widespread species, present all over the Mediterranean Sea from the Adriatic (Kovačić and Pallaoro, 2003) to the Aegean Sea (Ahnelt et al., 1995) (Fig. 4). Other Knipowitschia species have distributions restricted to a single drainage: three species are known in the Adriatic catchment, two species in the Ionian catchment,

Table II. - Degree of reduction of head canals and squamation: (a) all head canals developed; (b) the posterior oculoscapular canal absent; (c) the posterior oculoscapular and preopercular canals absent; (d) all canals absent; (1) scales along at least part of dorsal fins; (2) dorsal space naked; (3) squamation reduced to axillar and caudal peduncle patches; (4) squamation reduced to axillar patch; (1) reduction present in all specimens; (1) reduction of variable occurrence among specimens. [Degré de réduction des canaux céphaliques et des écailles: (a) tous les canaux céphaliques sont développés; (b) canal oculoscapulaire postérieur absent; (c) pas de canal oculoscapulaire postérieur ni préoperculaire; (d) aucun canal; (1) écailles au moins le long d'une partie des nageoires dorsales; (2) espace dorsal nu sans écaille; (3) écailles réduites à la zone axillaire et à la zone du pédoncule caudal; (4) écailles réduites à la zone axillaire; (1) réduction présente chez tous les spécimens; (11) réduction de l'occurrence variable parmi les spécimens.]



and three species in the Aegean catchment (Fig. 4, Tab. II). In addition, several undescribed *Knipowitschia* species were mentioned for the rivers Krka and Matica in the Adriatic catchment (Mrakovčić et al., 1996). The status of *K. panizzae* was recently questioned (Kovačić and Pallaoro, 2003). The theory on speciation of *Knipowitschia* in the Mediterranean region presumes that differentiation of *Knipowitschia* began during late Miocene salinity crisis (about 5 My) and the Lago Mare phase (4.9 My) (Ahnelt, 1995; Ahnelt et al., 1995). *K. caucasica* was the only species to enter the Mediterranean much later, during the melting phase after

the Würm glacial period (12,000-9,500 BP) (Ahnelt, 1995; Ahnelt et al., 1995). It is euryhaline species, like the other Black, Caspian and Aral Seas Knipowitschia species. This scenario explains restricted distribution of all Mediterranean Knipowitschia species, except K. caucasica. The reduction of head canals and the reduction of squamation of Mediterranean Knipowitschia is related to the reduction of body size, and explained by selection pressure for small and restricted habitats (Ahnelt, 1995). The intensity of these reductions differs within Knipowitschia (Tab. II). Similarities in squamation and head canals development were noted between some of these species: K. punctatissima and K. thessala (Economidis and Miller, 1990), and K. goerneri and K. mermere (Ahnelt, 1995) (Tab. II). Among known Knipowitschia species, K. radovici is morphologically closest to K. thessala. However, the reductions have a few basic degrees and the number of combinations is limited. Intraspecies variability is present within each degree of reduction, and sometimes more than one degree is present within the same species (Tab. II). These species live in similar habitats exposed to similar selection pressure. Therefore, similar reductions have probably been derived independently (Ahnelt, 1995), and are hardly results of closer relationship between geographically distant species. On the other hand, the reductions of squamation and head canals are useful characters for the identification of Knipowitschia species, especially among sympatric species (like Adriatic *K.* caucasica, *K.* punctatissima and *K*.

radovici). Other morphological characters (sensory papillae of lateral line system, fin meristics, body coloration) are of limited use for species diagnosis within *Knipowitschia*. Only a few characters of sensory papillae rows differ enough among *Knipowitschia* species to be used for species identification: Presence of rows p, m and u; development of transverse proliferation of row a; continuity of row z; anterior beginning of row b. The meristics of D2, A and P is even more useless, because it greatly overlaps among these species. Coloration in *Knipowitschia* differentiates more

between sexes than among species. The important difference in coloration between sexes is (1) Y shape mark on preorbital and chin area in females, (2) more distinctive and intensive spot in the rear part of D1 in males, (3) lateral vertical bars in males (not present in all Knipowitschia species), and (4) head and breast densely pigmented in males (possible seasonally dependent, Gandolfi et al., 1985). The coloration of females in all currently known Knipowitschia species is very similar (Ahnelt et al., 1995). The most distinguishing coloration characters in males among species are the presence, size and intensity of lateral vertical bars, and characteristics of D1 mark. In some species the description of coloration is incomplete: K. ephesi, K. mermere, K. punctatissima (Gandolfi et al., 1985; Ahnelt, 1991, 1995), or is complete for only one sex: K. milleri, K. radovici (Ahnelt and Bianco, 1990; present data).

Most Mediterranean *Knipowitschia* are endemic species, restricted to small areas. Furthermore, these endemic species live in isolated freshwater habitats vulnerable to human threats. Conservation of all these species is necessary to prevent extinction. The reasons for the survival of these species were elaborated by Miller (1990). However, our knowledge is insufficient for effective conservation of these species. Systematic observations of *Knipowitschia* populations of Mediterranean freshwaters are needed to provide necessary data for the conservation of these species, as it was accomplished for K. caucasica in Dalmatia (Kovačić and Pallaoro, 2003). Data should include: (1) Additional taxonomic characters for known species beneficial to the identification of species, and possible identification of new species. Some Knipowitschia species lack descriptive details, especially those described on the basis of old material in museum collections. Substantial sample of material could extend species taxonomical delimitation. The description of juvenile stages would aid species identification on further samples collected in months when juveniles dominate the populations of these short-living fishes. (2) Actual geographical distribution, based on systematically collected and correctly identified *Knipowitschia* samples. (3) Ranges of ecological conditions that are tolerated by the species, based on correctly identified samples and detailed habitat data for that sample. (4) Descriptions of new species that would possibly occur among systematically collected samples. (5) Species' biology, that could be researched on collected specimens.

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